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Research Article

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Scientific Evaluation of Ayurvedic Compound Formulation Dhanyapanchaka Kvatha Curna

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ABSTRACT

India is vast repository of medicinal plants that are used in traditional medical treatments. Herbal medicines as the major remedy in traditional system of medicine have been used in medical practices since antiquity. Quality assurance is an integral part of all systems of medicine to ensure quality medicament. Thus, there is an urgent need to evaluate such parameters which can be adopted by the pharmaceutical industries. To keep this view in mind, a polyherbal Dhanyapanchaka Kvatha Curna is formulated in house, which is very effective in Ãmaśũla (Pain); Ãmãtisãra (Diarrhoea due to indigestion) and Aruci (Tastelessness). The Kvatha curna formulated by five single ingredients viz. Dhãnyaka (Coriandrum sativum fruit.), Nãgara (Zingiber officinale Rhizome), Mustã (Cyperus rotundus Rhizome), Bãlaka (Coleus vettiveroides Root), Bilva (Aegle marmelos Fruit pulp) All the ingredients and kvatha curna were analyzed in order to assess the authenticity of the drugs based on ayurvedic requirment following a series of powder microscopy, physico-chemical, HPTLC finger printing and. The obtained values/ranges can adopted to pay down new pharmacopoeial standards, to be followed for traditional preparation of Dhanyapanchaka kvatha curna. And also can be used effectively for the identification of raw materials in the compound formulation.

Key words: *Dhanyapanchaka kvatha curna, Scientific evaluation, Physicochemical analysis, HPTLC finger print..*

INTRODUCTION

Standardized drugs of well defined consistent quality are needed for reliable beneficial therapeutic use. Total information and controls are necessary to guarantee consistency of composition. Due to lack of proper quality control methods, there are batch to batch variations in the same product as well as variations amongst the same product obtained from different sources. The main problem encountered while working with compound formulations is that most of them consist of several ingredients, and the presence of each ingredient has to be confirmed in the final product[1, 2].

To keep this view in mind, a polyherbal Dhanyapanchaka kvatha curna is formulated in house and Chitrakoot Ras Shala Pharmacy, Chitrakoot which is very effective in Ãmaśũla (Pain); Ãmãtisãra (Diarrhoea due to indigestion) and Aruci (Tastelessness). Its ingredients also widely used to cure various diseases and preparation of ayurvedic compound formulations[3, 4]. The kvatha curna formulated by five single ingredients *viz.* Dhãnyaka (*Coriandrum sativum* fruit.), Nãgara (*Zingiber officinale* rhizome), Mustã *(Cyperus rotundus rhizome*), Bãlaka (*Coleus vettiveroides* root), Bilva (*Aegle marmelos fruit pulp*). All the ingredients and kvatha curna were analyzed in order to assess the authenticity of the drugs based on ayurvedic requirment following a series of powder microscopy, physico-chemical, and HPTLC finger printing. The obtained values/ranges can adopted to pay down new pharmacopoeial standards, to be

followed for traditional preparation of Dhanyapanchaka kvatha Curna. And also can be used effectively for the identification of raw materials in the compound formulation.

MATERIAL AND METHODS

All the ingredients were used of pharmacopoeial quality[5]. These were cleaned, washed, dried and grinded individually, pass through 710 µm IS Sieve (old sieve number 22). Weighed separately and mixed them equal proportions (1:1:1:1:1) to ensure a homogenous mixture, these ware stored in an airtight containers to protect from light and moisture. Two samples were prepared at research laboratory Ayurveda Sadan, Chitrakoot Batch-A and Batch-B where Batch-C was prepared by Chitrakoot Ras-Shalsa Pharmacy.

Preparation of Slides for Microscopic Examination

For microscopic analysis about 2 gm of formulated kvatha curna in a small beaker and wash thoroughly with water, pour out the water without loss of material, mount a small portion in *glycerine*; warm a few mg with *chloral hydrate* solution, wash and mount in *glycerine*; treat a few mg with *iodine* in *potassium iodide* solution and mount in *glycerine*; treat a few mg with *phloroglucinol* and *conc. HCl* solution and mount in *glycerine*. about 0.5 g of sample and add 50 percent conc. *nitric acid* in a test tube and warm over water bath till brown fumes appear; wash with water thoroughly and mount a small portion in *glycerin*. Observed the characteristics in the various mounts[6,7].

Physicochemical Tests

Organoleptic characters, particle size and physic-chemical analysis of all the samples were carried out. Quantitative analysis for loss on drying at 105^{o}C, alcohol soluble extractive, water soluble extractive, total ash, acid insoluble ash[8].

High Performance Thin Layer Chromatography (HPTLC) Profile

For HPTLC, 2gm of each sample was extracted with 25 ml of methanol on boiling water bath for 25 minute consecutively of 3 times using fresh portion of 25 ml methanol, filtrate and concentrated. TLC of extracts of all the samples was carried out on Silica Gel 60 F_{254} precoated plates (0.2 mm thickness; from Merck India Limited Mumbali). An applicator from Camag Linomat-5 (Camag Switzerland 140443) was used for band application and photo documentation unit (Camag Reprostar-3: 140604) was used for documentation of chromatographic fingerprints. The mobile phase used *Toluene: Ethyl acetate* (6:4). The plate was developed over a distance of 9 cm in a saturated development chamber (Twin trough chamber (10 x 10 cm with SS lid, and visualized under visible light, 254nm and 366nm. After spraying with 5% methanolic sulphuric acid followed by heating at 110^{0}C for 5-10 minute[9,10].

RESULTS AND DISCUSSION

Description

A coarse powder, light brown in colour with odour of ginger, taste slightly bitter. All particles pass through 710 µm IS Sieve (old sieve number 22) and not more than 10 percent passes through 355 µm IS Sieve (old sieve number 22).

Physicochemical parameters

Physicochemical tests were done and results are given in table -1

Powder microscopic characters

Epicarp fragments with thick and straight walled polygonal cells containing prisms of calcium-oxalate crystals in surface view, mesocarpic sclerified layers of fibres crisscrossing each other of right angles, no trichomes or vittae debris (Dhãnyaka Fig. A, B, C); parenchymatous cells containing oleo-resin, oval to round starch grains not less than 15µ to 30µ and several up to 70µ with hilum ecentric, lamellae distinct, pitted, septate fibres with indentations on its walls (Nãgara Fig. D, E, F); fibre sclereids from scale leaves in packed rows, beak shaped starch grains 6µ to 28µ, narrow vessel with scalariform thickening (Mustã Fig. G, H, I); cork cells in surface view, fibre tracheids showing branching or splitting at the tips, reticulately marked parenchymatous cells (Bãlaka Fig. J, K, L); thick walled round to oval elongated parenchymatous cells containing oil globules and small prisms of calcium oxalate crystals, groups of

round to oval stone cells with large lumen, elongated , pitted sclereids, endosperm cells filled with oil globules, prismatic crystals of calcium oxalate, simple and compound starch grains, testa in surface view, fibres (Bilva Fig. M, N, O, P, Q, R).

HPTLC finger print profile

High performance thin layer chromatography (HPTLC) study of the methanolic extract 3 spots of the sample extracts applied in the TLC plate. Major spots R_f values with colour were recorded under 254nm, 366nm, after derivatization 366nm and Visible light. Chromatogram profile and R_f values are given (Plate 3, Fig. S, T, U, V & Table 2-5).

The powder microscopic diagnostic features, physicochemical tests, have been established to identify and strength of *Dhanyapanchaka kvatha curna*. The microscopical parameters can be used for checking the adulteration and purity of this compound formulation. HPTLC finger print profile helps in identification of various ingredients present in the Dhanyapanchaka kvatha curna thereby substantiating and authenticating of crude drug. These finding could be helpful in identification and authentication.

Table 1: Physico-chemical parameters of Dhãnyapañchaka Kvãtha cũrṇa and ingredients

Name of Curna	LOD (%w/w)	Total ash (% w/w)	AI ash (% w/w)	ASE (% w/w)	WSE (% w/w)
Dhãnyapañchaka Kvãtha cũrṇa (Batch A)	6.31	4.55	1.66	4.37	15.50
Dhãnyapañchaka Kvãtha cũrṇa (Batch B)	6.06	4.67	1.73	5.08	15.99
Dhãnyapañchaka Kvãtha cũrṇa (Batch C)	7.01	5.01	1.86	4.64	15.54
Dhãnyaka	5.79	4.69	0.94	23.55	22.22
Nãgara (Sunthî)	8.68	5.84	0. 97	4.14	15.45
Musta (Mustã)	7.54	2.87	1.86	9.78	17.40
Bãlaka (hrîvera)	6.44	7.92	2.36	30.36	23.02
Bilva	8.17	1.78	0.79	18.66	73.22

Table 2: R_f values in TLC finger prints of Dhãnyapañchaka Kvãtha cũrṇa at 254 nm

R_f values	Dhãnyapañchaka Kvãtha cũrṇa		
	Batch 01	**Batch 02**	**Batch 03**
R_f 1 (black)	0.20	0.20	0.20
R_f 2 (black)	0.44	0.44	0.44
R_f 3 (black)	0.78	0.78	0.78
R_f 4 (black)	0.88	0.88	0.88

Table 3: R_f values in TLC finger prints of Dhãnyapañchaka Kvãtha cũrṇa at 366 nm

R_f values	Dhãnyapañchaka Kvãtha cũrṇa		
	Batch 01	**Batch 02**	**Batch 03**
R_f 1 (blue)	0.16	0.16	0.16
R_f 2 (blue)	0.27	0.27	0.27
R_f 3 (white)	0.39	0.39	0.39
R_f 4 (blue)	0.59	0.59	0.59
R_f 5 (brown)	0.74	0.74	0.74
R_f 6 (sky blue)	0.87	0.87	0.87
R_f 7 (pink)	0.93	0.93	0.93

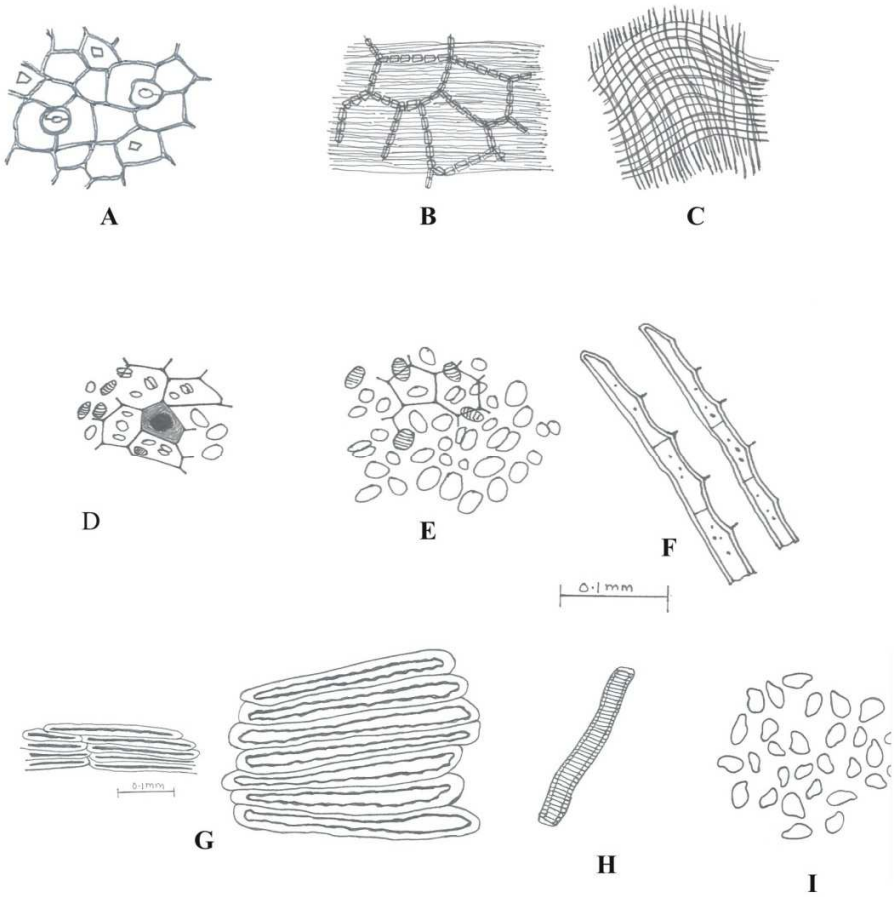
Table 4: R_f values in TLC finger prints of Dhãnyapañchaka Kvãtha cũrṇa at 366 nm (after derivatization)

Rf values	Dhãnyapañchaka Kvãtha cũrṇa		
	Batch 01	Batch 02	Batch 03
R_f1(brown)	0.11	0.11	0.11
R_f 2 (blue)	0.17	0.17	0.17
R_f 3 (brown)	0.33	0.33	0.33
R_f4(brown)	0.39	0.39	0.39
R_f5(white)	0.66	0.66	0.66
R_f6(sky blue)	0.78	0.78	0.78
R_f 7 (light blue)	0.87	0.87	0.87

Table 5: R_f values in TLC finger prints of Dhãnyapañchaka Kvãtha cũrṇa at visible light (after derivatization)

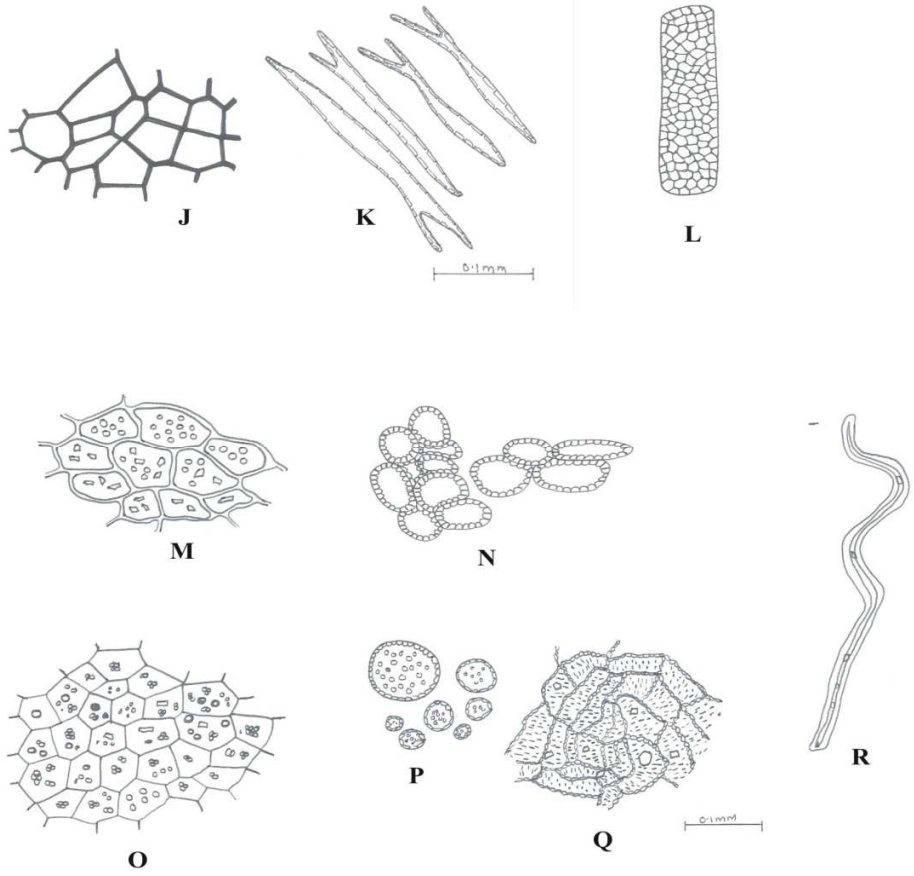
R_f values	Dhãnyapañchaka Kvãtha cũrṇa		
	Batch 01	Batch 02	Batch 03
R_f 1 (brown)	0.11	0.11	0.11
R_f 2 (brown)	0.30	0.30	0.30
R_f 3 (brown)	0.46	0.46	0.46
R_f 4 (brown)	0.72	0.72	0.72
R_f 5 (black)	0.93	0.93	0.93

Plate1: Microscopic characters of Dhãnyapañchaka Kvãtha cũrṇa



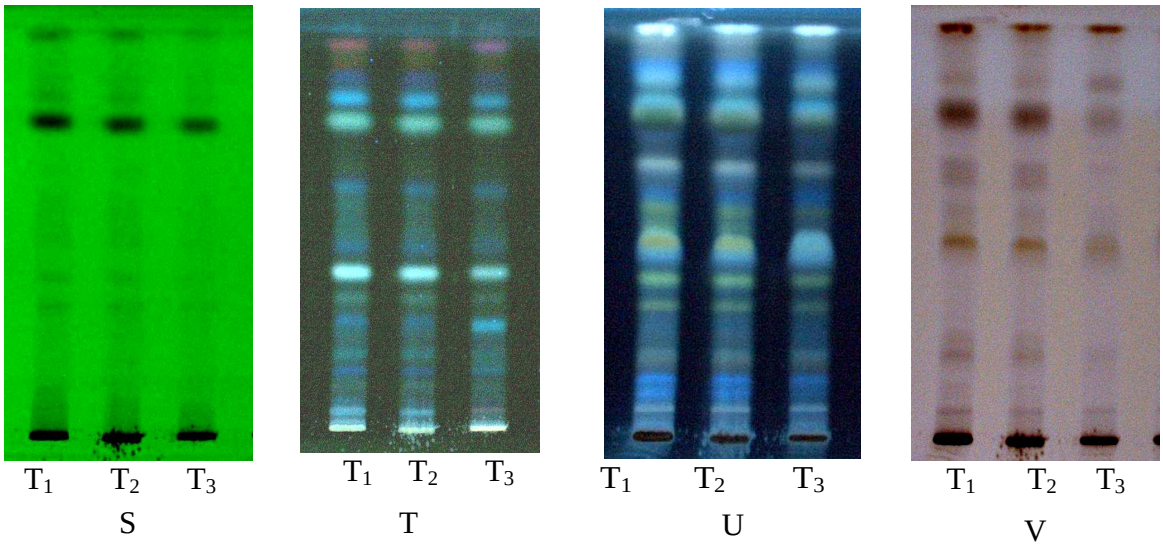
Dhãnyaka A:- Fragments of epicarp, B:- Endocarpic layer and pitted parenchyma, C: Crisscross fibres; Nãgara D:- Parenchymatous cells containing oleo-resin, E:- Starch grains, F:- Pitted septate fibres; Mustã G: Fibre - sclerids from scale leaves cells, H:- Scalariform vessel, I:- starch grains

Plate2: Microscopic characters of Dhãnyapañchaka Kvãtha cũrṇa



Balaka J:- Cork cells in surface view, K:- Branched fibres tracheids, L:- Reticulate parenchyma; Bilva M:- Parenchymatous cells containing oil globules and calcium oxalate crystals in surface view, N:- Groups of oval stone cells, O:- Endosperm cell with starch grains, crystals and oil globules, P:- Oil globules, Q:- Testa in surface

Plate 3: TLC finger prints of test solution of Dhãnyapañchaka Kvãth



Track 1: Batch 01, Track 2: Batch 02 and Track 3: Batch 03

S:- TLC profile of Dhãnyapañchaka Kvãtha cũrṇa observed under 254nm, T:- TLC profile of Dhãnyapañchaka Kvãtha cũrṇa observed under 366nm, U:- TLC profile of Dhãnyapañchaka Kvãtha cũrṇa after spraying with 5 % *methanolic sulphuric acid* reagent reagent observed under 366nm, V:- TLC profile of Dhãnyapañchaka Kvãtha cũrṇa after spraying with 5 % *methanolic sulphuric acid* reagent observed under visible light

CONCLUSION

Dhanyapanchaka kvatha curna and its ingredients have numerous uses in Ayurvedic medicine and traditional medicine to treat several ailments like used in Ãmaśũla (Pain); Ãmãtisãra (Diarrhoea due to indigestion) and Aruci (Tastelessness). Thus, the present study revealed that the microbiological assessment, physicochemical tests, characteristics microscopical features and the distinguished finger prints in the HPTLC profiles may be utilized as marker parameters for monitoring the quality of the formulation. All the parameters may be also used for quality evaluation and the standardization of compound formulations. Also, standardization and development for reliable quality protocols for ayurvedic formulations are important for keeping a check on batch to batch variations. Hence, the physicochemical parameters, quantitative analysis, microbiological assessment, HPTLC fingerprinting profiles and the microscopic characteristics together may be used for quality evaluation and the standardization of compound formulations and maintaining their quality, purity and efficacy.

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